

(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau



(43) International Publication Date
4 August 2005 (04.08.2005)

PCT

(10) International Publication Number
WO 2005/070122 A3

(51) International Patent Classification⁷: C12Q 1/00

(21) International Application Number:
PCT/US2005/000722

(22) International Filing Date: 10 January 2005 (10.01.2005)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
60/535,327 9 January 2004 (09.01.2004) US

(71) Applicant (for all designated States except US): **BOARDS OF REGENTS, THE UNIVERSITY OF TEXAS SYSTEM** [US/US]; 201 West 7th Street, Austin, TX 78701 (US).

(72) Inventors; and

(75) Inventors/Applicants (for US only): **HAMILL, Owen, P.** [US/US]; 1212 Winnie Street, Galveston, TX 77550 (US). **MAROTO, Rosario** [ES/US]; 1212 Winnie Street, Galveston, TX 77550 (US).

(74) Agent: **PROVENCE, David, L.**; Muetting, Raasch & Gebhardt, P.A., P.O. Box 581415, Minneapolis, MN 55458-1415 (US).

(81) Designated States (unless otherwise indicated, for every kind of national protection available): AE, AG, AL, AM,

AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SM, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW.

(84) Designated States (unless otherwise indicated, for every kind of regional protection available): ARIPO (BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IS, IT, LT, LU, MC, NL, PL, PT, RO, SE, SI, SK, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:

- with international search report
- before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments

(88) Date of publication of the international search report:
16 March 2006

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

(54) Title: MECHANOSENSITIVE ION CHANNELS AND METHODS OF USE

(57) Abstract: The present invention provides methods for identifying agents that decrease the activity of a mechanosensitive ion channels, preferably, a mechanosensitive Ca²⁺-permeable channel (MscCa) channel. The present invention also provides methods for using agents that decrease the activity of mechanosensitive ion channels, including, for instance, methods for treating cancer, methods for decreasing metastasis of a cancer cell, and methods for decreasing a symptom associated with cancer.



WO 2005/070122 A3

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US05/00722

A. CLASSIFICATION OF SUBJECT MATTER IPC(7) : C12Q 1/00 US CL : 435/4 According to International Patent Classification (IPC) or to both national classification and IPC																				
B. FIELDS SEARCHED Minimum documentation searched (classification system followed by classification symbols) U.S. : 435/4 Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched Electronic data base consulted during the international search (name of data base and, where practicable, search terms used) medline, cancerlit, biosis, uspatents																				
C. DOCUMENTS CONSIDERED TO BE RELEVANT																				
Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.																		
X	LEE et al. Regulation of cell movement is mediated by stretch-activated calcium channels. Nature. July 1999, Vol. 400, pages 382-386.	1,7																		
X	YAO et al. A protein kinase G-sensitive channel mediates flow-induced Ca ²⁺ entry into vascular endothelial cells. FASEB Journal. May 2000, Vol. 14, pages 932-938.	1																		
<input type="checkbox"/> Further documents are listed in the continuation of Box C. <input type="checkbox"/> See patent family annex.																				
<table border="0"><tr><td>* Special categories of cited documents:</td><td>"T"</td><td>later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention</td></tr><tr><td>"A" document defining the general state of the art which is not considered to be of particular relevance</td><td>"X"</td><td>document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone</td></tr><tr><td>"E" earlier application or patent published on or after the international filing date</td><td>"Y"</td><td>document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art</td></tr><tr><td>"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)</td><td>"&"</td><td>document member of the same patent family</td></tr><tr><td>"O" document referring to an oral disclosure, use, exhibition or other means</td><td></td><td></td></tr><tr><td>"P" document published prior to the international filing date but later than the priority date claimed</td><td></td><td></td></tr></table>			* Special categories of cited documents:	"T"	later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention	"A" document defining the general state of the art which is not considered to be of particular relevance	"X"	document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone	"E" earlier application or patent published on or after the international filing date	"Y"	document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art	"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)	"&"	document member of the same patent family	"O" document referring to an oral disclosure, use, exhibition or other means			"P" document published prior to the international filing date but later than the priority date claimed		
* Special categories of cited documents:	"T"	later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention																		
"A" document defining the general state of the art which is not considered to be of particular relevance	"X"	document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone																		
"E" earlier application or patent published on or after the international filing date	"Y"	document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art																		
"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)	"&"	document member of the same patent family																		
"O" document referring to an oral disclosure, use, exhibition or other means																				
"P" document published prior to the international filing date but later than the priority date claimed																				
Date of the actual completion of the international search 01 December 2005 (01.12.2005)		Date of mailing of the international search report 26 JAN 2006																		
Name and mailing address of the ISA/US Mail Stop PCT, Attn: ISA/US Commissioner for Patents P.O. Box 1450 Alexandria, Virginia 22313-1450 Facsimile No. (571) 273-3201		Authorized officer Gary B. Nickol Ph.D. <i>Janice Ford</i> Telephone No. 703-308-0196																		

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US05/00722

Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:
2. ☐ Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:
3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:
Please See Continuation Sheet

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying additional fees, this Authority did not invite payment of any additional fees.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. ☒ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.: 1 and 7

- Remark on Protest
- ☐ The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.
 - ☐ The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.
 - ☐ No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US05/00722

BOX III. OBSERVATIONS WHERE UNITY OF INVENTION IS LACKING

This application contains the following inventions or groups of inventions which are not so linked as to form a single general inventive concept under PCT Rule 13.1. In order for all inventions to be examined, the appropriate additional examination fees must be paid.

Group 1, claim(s) 1, 7, drawn to the special technical feature for identifying an agent that decreases activity of a mechanosensitive Ca²⁺ channel comprising contacting a motile cell expressing said channel with a candidate agent.

Group 2, claim(s) 1-6, 8-10, drawn to the special technical feature for identifying an agent that decreases activity of a mechanosensitive Ca²⁺ channel (MscCa) comprising contacting a tumor cell expressing the MscCa channel wherein said channel comprises SEQ ID NO:2.

Group 3, claim(s) 11, drawn to the special technical feature of an agent that decreases activity of a MscCa channel.

Group 4, claim(s) 12-18, drawn to the special technical feature for identifying an agent that decreases a phenotype of a cell comprising contacting an MscCa channel with a candidate agent to yield a treated cell.

Group 5, claim(s) 19, drawn to the special technical feature of an agent that decreases the phenotype of a cell that expresses an MscCa channel.

Group 6, claim(s) 20-25, 29-30, drawn to the special technical feature of a method for treating cancer comprising administering a polypeptide agent that decreases the activity of a mechanosensitive ion channel present on cancer cells.

Group 7, claim(s) 20-23, 26-27, 29-30, drawn to the special technical feature of a method for treating cancer comprising administering an antibody that decreases the activity of a mechanosensitive ion channel present on cancer cells.

Group 8, claim(s) 20-23, 28-34, drawn to the special technical feature of a method for treating cancer comprising administering a polynucleotide that decreases expression of a MscCa polypeptide.

The inventions listed as Groups 1-8 do not relate to a single general inventive concept under PCT Rule 13.1 because, under PCT Rule 13.2, they lack the same or corresponding special technical features for the following reasons:

The technical feature linking Groups 1-8 appears to be a method for identifying an agent that decreases activity of a mechanosensitive Ca²⁺ permeable (MscCa) channel comprising contacting a cell expressing an MscCa channel with a candidate agent wherein decreased activity of an MscCa channel indicates that the candidate agent decreases the activity of an MscCa channel.

However, Yao *et al.* (A protein kinase G-sensitive channel mediates flow-induced Ca²⁺ entry into vascular endothelial cells, FASEB Journal, May 2000, Vol. 14, pages 932-938) identify a mechanosensitive Ca²⁺ permeable cation channel and teach that inhibition of this channel abolished the rise of calcium ions (page 932, second column, last paragraph). Yao *et al.* further identify agents that decrease the activity of said channels in cells compared to controls (page 936, second column).

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US05/00722

Therefore, the technical feature linking the inventions of Groups 1-8 does not constitute a special technical feature as defined by PCT Rule 13.2 as it does not define a contribution over the prior art.

PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY (Chapter I of the Patent Cooperation Treaty)

(PCT Rule 44bis)

Applicant's or agent's file reference 265.00450201	FOR FURTHER ACTION		See item 4 below
International application No. PCT/US2005/000722	International filing date (<i>day/month/year</i>) 10 January 2005 (10.01.2005)	Priority date (<i>day/month/year</i>) 09 January 2004 (09.01.2004)	
International Patent Classification (8th edition unless older edition indicated) See relevant information in Form PCT/ISA/237			
Applicant BOARDS OF REGENTS, THE UNIVERSITY OF TEXAS SYSTEM			

1. This international preliminary report on patentability (Chapter I) is issued by the International Bureau on behalf of the International Searching Authority under Rule 44 bis.1(a).

2. This REPORT consists of a total of 5 sheets, including this cover sheet.

In the attached sheets, any reference to the written opinion of the International Searching Authority should be read as a reference to the international preliminary report on patentability (Chapter I) instead.

3. This report contains indications relating to the following items:

- | | |
|---|---|
| <input checked="" type="checkbox"/> Box No. I | Basis of the report |
| <input type="checkbox"/> Box No. II | Priority |
| <input checked="" type="checkbox"/> Box No. III | Non-establishment of opinion with regard to novelty, inventive step and industrial applicability |
| <input type="checkbox"/> Box No. IV | Lack of unity of invention |
| <input checked="" type="checkbox"/> Box No. V | Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement |
| <input type="checkbox"/> Box No. VI | Certain documents cited |
| <input type="checkbox"/> Box No. VII | Certain defects in the international application |
| <input type="checkbox"/> Box No. VIII | Certain observations on the international application |

4. The International Bureau will communicate this report to designated Offices in accordance with Rules 44bis.3(c) and 93bis.1 but not, except where the applicant makes an express request under Article 23(2), before the expiration of 30 months from the priority date (Rule 44bis .2).

The International Bureau of WIPO 34, chemin des Colombettes 1211 Geneva 20, Switzerland		Date of issuance of this report 10 July 2006 (10.07.2006)
Facsimile No. +41 22 338 82 70		Authorized officer Simin Baharlou
Form PCT/IB/373 (January 2004)		e-mail: pt09@wipo.int

From the
INTERNATIONAL SEARCHING AUTHORITY

PATENT COOPERATION TREATY

To:
DAVID L. PROVENCE
MUETING, RAASCH & GEBHARDT, P.A.
P.O. BOX 581415
MINNEAPOLIS, MN 55458-1415

REC'D 30 JAN 2006

WIPO PCT

WRITTEN OPINION OF THE
INTERNATIONAL SEARCHING AUTHORITY

(PCT Rule 43bis.1)

Applicant's or agent's file reference 265.00450201		Date of mailing (day/month/year) 26 JAN 2006
FOR FURTHER ACTION See paragraph 2 below		
International application No. PCT/US05/00722	International filing date (day/month/year) 10 January 2005 (10.01.2005)	Priority date (day/month/year) 09 January 2004 (09.01.2004)
International Patent Classification (IPC) or both national classification and IPC IPC(7): C12Q 1/00 and US Cl.: 435/4		
Applicant BOARD OF REGENTS, THE UNIVERSITY OF TEXAS SYSTEM		

1. This opinion contains indications relating to the following items:

- | | | |
|-------------------------------------|--------------|--|
| <input checked="" type="checkbox"/> | Box No. I | Basis of the opinion |
| <input type="checkbox"/> | Box No. II | Priority |
| <input checked="" type="checkbox"/> | Box No. III | Non-establishment of opinion with regard to novelty, inventive step and industrial applicability |
| <input type="checkbox"/> | Box No. IV | Lack of unity of invention |
| <input checked="" type="checkbox"/> | Box No. V | Reasoned statement under Rule 43bis.1(a)(i) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement |
| <input type="checkbox"/> | Box No. VI | Certain documents cited |
| <input type="checkbox"/> | Box No. VII | Certain defects in the international application |
| <input type="checkbox"/> | Box No. VIII | Certain observations on the international application |

2. FURTHER ACTION

If a demand for international preliminary examination is made, this opinion will be considered to be a written opinion of the International Preliminary Examining Authority ("IPEA") except that this does not apply where the applicant chooses an Authority other than this one to be the IPEA and the chosen IPEA has notified the International Bureau under Rule 66.1bis(b) that written opinions of this International Searching Authority will not be so considered.

If this opinion is, as provided above, considered to be a written opinion of the IPEA, the applicant is invited to submit to the IPEA a written reply together, where appropriate, with amendments, before the expiration of 3 months from the date of mailing of Form PCT/ISA/220 or before the expiration of 22 months from the priority date, whichever expires later.

For further options, see Form PCT/ISA/220.

3. For further details, see notes to Form PCT/ISA/220.

Name and mailing address of the ISA/ US Mail Stop PCT, Attn: ISA/US Commissioner for Patents P.O. Box 1450 Alexandria, Virginia 22313-1450 Facsimile No. (571) 273-3201	Date of completion of this opinion 01 December 2005 (01.12.2005)	Authorized officer Gary B. Nickol Ph.D. <i>Janice Ford</i> Telephone No. 703-308-0196
--	---	---

Form PCT/ISA/237 (cover sheet) (April 2005)

WRITTEN OPINION OF THE
INTERNATIONAL SEARCHING AUTHORITY

International application No.

PCT/US05/00722

Box No. I Basis of this opinion

1. With regard to the language, this opinion has been established on the basis of:
- ☒ the international application in the language in which it was filed
- ☐ a translation of the international application into _____, which is the language of a translation furnished for the purposes of international search (Rules 12.3(a) and 23.1(b)).
2. With regard to any nucleotide and/or amino acid sequence disclosed in the international application and necessary to the claimed invention, this opinion has been established on the basis of:
- a. type of material
- ☐ a sequence listing
- ☐ table(s) related to the sequence listing
- b. format of material
- ☐ on paper
- ☐ in electronic form
- c. time of filing/furnishing
- ☐ contained in the international application as filed.
- ☐ filed together with the international application in electronic form.
- ☐ furnished subsequently to this Authority for the purposes of search.
3. ☐ In addition, in the case that more than one version or copy of a sequence listing and/or table(s) relating thereto has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.
4. Additional comments:

WRITTEN OPINION OF THE
INTERNATIONAL SEARCHING AUTHORITY

International application No.

PCT/US05/00722

Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

The questions whether the claimed invention appears to be novel, to involve an inventive step (to be non-obvious), or to be industrially applicable have not been examined in respect of:

☐ the entire international application

☒ claims Nos. 2-6 and 8-34

because:

☐ the said international application, or the said claim Nos. _____ relate to the following subject matter which does not require an international search (*specify*):

☐ the description, claims or drawings (*indicate particular elements below*) or said claims Nos. _____ are so unclear that no meaningful opinion could be formed (*specify*):

☐ the claims, or said claims Nos. _____ are so inadequately supported by the description that no meaningful opinion could be formed (*specify*):

☒ no international search report has been established for said claims Nos. 2-6 and 8-34

☐ a meaningful opinion could not be formed without the sequence listing, the applicant did not, within the prescribed time limit:

☐ furnish a sequence listing on paper complying with the standard provided for in Annex C of the Administrative Instructions, and such listing was not available to the International Searching Authority in a form and manner acceptable to it.

☐ furnish a sequence listing in electronic form complying with the standard provided for in Annex C of the Administrative Instructions, and such listing was not available to the International Searching Authority in a form and manner acceptable to it.

☐ pay the required late furnishing fee for the furnishing of a sequence listing in response to an invitation under Rules 13ter.1(a) or (b).

☐ a meaningful opinion could not be formed without the tables related to the sequence listings; the applicant did not, within the prescribed time limit, furnish such tables in electronic form complying with the technical requirements provided for in Annex C-bis of the Administrative Instructions, and such tables were not available to the International Searching Authority in a form and manner acceptable to it.

☐ the tables related to the nucleotide and/or amino acid sequence listing, if in electronic form only, do not comply with the technical requirements provided for in Annex C-bis of the Administrative Instructions.

☐ See Supplemental Box for further details.

WRITTEN OPINION OF THE
INTERNATIONAL SEARCHING AUTHORITY

International application No.
PCT/US05/00722

Box No. V Reasoned statement under Rule 43 bis.1(a)(i) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)

Claims NONE YES
Claims 1, 7 NO

Inventive step (IS)

Claims NONE YES
Claims 1, 7 NO

Industrial applicability (IA)

Claims 1, 7 YES
Claims NONE NO

2. Citations and explanations:

Claims 1, 7 lack novelty under PCT Article 33(2) as being anticipated by LEE *et al.* "Regulation of cell movement is mediated by stretch-activated calcium channel" Nature. July 1999. Vol. 400, pages 382-386.

Lee *et al.* teach the identity of an agent (gadolinium- see page 384, 2nd column) that decreases the activity of a mechanosensitive Ca²⁺ channel in a motile cell wherein the treated motile cell has decreased motility compared to controls (see Figure 3b). The decreased motility was observed in fish keratocytes, which are capable of rapid locomotion (page 382, 1st paragraph). The reference teaches (page 384, 2nd column, 1st paragraph) that gadolinium treatment inhibited detachment of the rear cell margin, causing cells to become elongated and eventually to cease movement.

Claim 1 lacks novelty under PCT Article 33(2) as being anticipated by YAO *et al.* (A protein kinase G-sensitive channel mediates flow-induced Ca²⁺ entry into vascular endothelial cells, FASEB Journal, May 2000, Vol. 14, pages 932-938).

Yao *et al.* identify a mechanosensitive Ca²⁺ permeable cation channel and teach that inhibition of this channel abolished the rise of calcium ions (page 932, second column, last paragraph). Yao *et al.* further identify agents that decrease the activity of said channels in cells compared to controls (page 936, second column).

Claims 1,7 meet the criteria set out in PCT Article 33(4), and thus meets industrial applicability because the subject matter claimed can be made or used in industry.

**This Page is Inserted by IFW Indexing and Scanning
Operations and is not part of the Official Record.**

BEST AVAILABLE IMAGES

Defective images within this document are accurate representations of the original documents submitted by the applicant.

Defects in the images include but are not limited to the items checked:

- ☐ BLACK BORDERS
- ☐ IMAGE CUT OFF AT TOP, BOTTOM OR SIDES
- ☐ FADED TEXT OR DRAWING
- ☐ BLURRED OR ILLEGIBLE TEXT OR DRAWING
- ☐ SKEWED/SLANTED IMAGES
- ☐ COLOR OR BLACK AND WHITE PHOTOGRAPHS
- ☐ GRAY SCALE DOCUMENTS
- ☐ LINES OR MARKS ON ORIGINAL DOCUMENT
- ☒ REFERENCE(S) OR EXHIBIT(S) SUBMITTED ARE POOR QUALITY
- ☐ OTHER: _____

IMAGES ARE BEST AVAILABLE COPY.

As rescanning these documents will not correct the image problems checked, please do not report these problems to the IFW Image Problem Mailbox.